



## SEQUENCE LISTING

<110> Short, Jay M.  
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Garrett, James B.  
O' Donoghue, Eileen  
Mathur, Eric J.

<120> RECOMBINANT BACTERIAL PHYTASES AND USES  
THEREOF

<130> 09010-029006

<140> US 09/866,379

<141> 2001-05-24

<150> US 09/580,515

<151> 2000-05-25

<150> US 09/318,528

<151> 1999-05-25

<150> US 09/291,931

<151> 1999-04-13

<150> US 09/259,214

<151> 1999-03-01

<150> US 08/910,798

<151> 1997-08-13

<160> 10

<170> FastSEQ for Windows Version 4.0

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<211> 1323

<212> DNA

<213> Escherichia coli

<220>

<221> CDS

<222> (1)...(1320)

<221> misc\_feature

<222> 216

<223> n = A,T,C or G

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1 5 10 15

ccg caa tct gca ttc gct cag agt gag ccg gag ctg aag ctg gaa agt 96

Pro	Gln	Ser	Ala	Phe	Ala	Gln	Ser	Glu	Pro	Glu	Leu	Lys	Leu	Glu	Ser	
			20					25					30			
gtg	gtg	att	gtc	agt	cgt	cat	ggg	gtg	cgt	gct	cca	acc	aag	gcc	acg	144
Val	Val	Ile	Val	Ser	Arg	His	Gly	Val	Arg	Ala	Pro	Thr	Lys	Ala	Thr	
		35					40					45				
caa	ctg	atg	cag	gat	gtc	acc	cca	gac	gca	tgg	cca	acc	tgg	ccg	gta	192
Gln	Leu	Met	Gln	Asp	Val	Thr	Pro	Asp	Ala	Trp	Pro	Thr	Trp	Pro	Val	
	50					55					60					
aaa	ctg	ggg	tgg	ctg	aca	ccg	cgn	ggg	ggg	gag	cta	atc	gcc	tat	ctc	240
Lys	Leu	Gly	Trp	Leu	Thr	Pro	Arg	Gly	Gly	Glu	Leu	Ile	Ala	Tyr	Leu	
	65				70				75						80	
gga	cat	tac	caa	cgc	cag	cgt	ctg	gta	gcc	gac	gga	ttg	ctg	gcg	aaa	288
Gly	His	Tyr	Gln	Arg	Gln	Arg	Leu	Val	Ala	Asp	Gly	Leu	Leu	Ala	Lys	
			85					90						95		
aag	ggc	tgc	ccg	cag	tct	ggg	cag	gtc	gcg	att	att	gct	gat	gtc	gac	336
Lys	Gly	Cys	Pro	Gln	Ser	Gly	Gln	Val	Ala	Ile	Ile	Ala	Asp	Val	Asp	
			100					105					110			
gag	cgt	acc	cgt	aaa	aca	ggc	gaa	gcc	ttc	gcc	gcc	ggg	ctg	gca	cct	384
Glu	Arg	Thr	Arg	Lys	Thr	Gly	Glu	Ala	Phe	Ala	Ala	Gly	Leu	Ala	Pro	
		115					120					125				
gac	tgt	gca	ata	acc	gta	cat	acc	cag	gca	gat	acg	tcc	agt	ccc	gat	432
Asp	Cys	Ala	Ile	Thr	Val	His	Thr	Gln	Ala	Asp	Thr	Ser	Ser	Pro	Asp	
	130					135					140					
ccg	tta	ttt	aat	cct	cta	aaa	act	ggc	gtt	tgc	caa	ctg	gat	aac	gcg	480
Pro	Leu	Phe	Asn	Pro	Leu	Lys	Thr	Gly	Val	Cys	Gln	Leu	Asp	Asn	Ala	
	145				150					155					160	
aac	gtg	act	gac	gcg	atc	ctc	agc	agg	gca	gga	ggg	tca	att	gct	gac	528
Asn	Val	Thr	Asp	Ala	Ile	Leu	Ser	Arg	Ala	Gly	Gly	Ser	Ile	Ala	Asp	
				165					170					175		
ttt	acc	ggg	cat	cgg	caa	acg	gcg	ttt	cgc	gaa	ctg	gaa	cgg	gtg	ctt	576
Phe	Thr	Gly	His	Arg	Gln	Thr	Ala	Phe	Arg	Glu	Leu	Glu	Arg	Val	Leu	
			180					185						190		
aat	ttt	ccg	caa	tca	aac	ttg	tgc	ctt	aaa	cgt	gag	aaa	cag	gac	gaa	624
Asn	Phe	Pro	Gln	Ser	Asn	Leu	Cys	Leu	Lys	Arg	Glu	Lys	Gln	Asp	Glu	
		195					200					205				
agc	tgt	tca	tta	acg	cag	gca	tta	cca	tcg	gaa	ctc	aag	gtg	agc	gcc	672
Ser	Cys	Ser	Leu	Thr	Gln	Ala	Leu	Pro	Ser	Glu	Leu	Lys	Val	Ser	Ala	
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gac	aat	gtc	tca	tta	acc	ggg	gcg	gta	agc	ctc	gca	tca	atg	ctg	acg	720
Asp	Asn	Val	Ser	Leu	Thr	Gly	Ala	Val	Ser	Leu	Ala	Ser	Met	Leu	Thr	
	225					230				235					240	
gag	ata	ttt	ctc	ctg	caa	caa	gca	cag	gga	atg	ccg	gag	ccg	ggg	tgg	768
Glu	Ile	Phe	Leu	Leu	Gln	Gln	Ala	Gln	Gly	Met	Pro	Glu	Pro	Gly	Trp	

245	250	255	
gga agg atc acc gat tca cac cag tgg aac acc ttg cta agt ttg cat			816
Gly Arg Ile Thr Asp Ser His Gln Trp Asn Thr Leu Leu Ser Leu His			
260	265	270	
aac gcg caa ttt tat ttg cta caa cgc acg cca gag gtt gcc cgc agc			864
Asn Ala Gln Phe Tyr Leu Leu Gln Arg Thr Pro Glu Val Ala Arg Ser			
275	280	285	
cgc gcc acc ccg tta ttg gat ttg atc atg gca gcg ttg acg ccc cat			912
Arg Ala Thr Pro Leu Leu Asp Leu Ile Met Ala Ala Leu Thr Pro His			
290	295	300	
cca ccg caa aaa cag gcg tat ggt gtg aca tta ccc act tca gta ctg			960
Pro Pro Gln Lys Gln Ala Tyr Gly Val Thr Leu Pro Thr Ser Val Leu			
305	310	315	320
ttt att gcc gga cac gat act aat ctg gca aat ctc ggc ggc gca ctg			1008
Phe Ile Ala Gly His Asp Thr Asn Leu Ala Asn Leu Gly Gly Ala Leu			
325	330	335	
gag ctc aac tgg acg ctt ccc ggt cag ccg gat aac acg ccg cca ggt			1056
Glu Leu Asn Trp Thr Leu Pro Gly Gln Pro Asp Asn Thr Pro Pro Gly			
340	345	350	
ggt gaa ctg gtg ttt gaa cgc tgg cgt cgg cta agc gat aac agc cag			1104
Gly Glu Leu Val Phe Glu Arg Trp Arg Arg Leu Ser Asp Asn Ser Gln			
355	360	365	
tgg att cag gtt tcg ctg gtc ttc cag act tta cag cag atg cgt gat			1152
Trp Ile Gln Val Ser Leu Val Phe Gln Thr Leu Gln Gln Met Arg Asp			
370	375	380	
aaa acg ccg ctg tca tta aat acg ccg ccc gga gag gtg aaa ctg acc			1200
Lys Thr Pro Leu Ser Leu Asn Thr Pro Pro Gly Glu Val Lys Leu Thr			
385	390	395	400
ctg gca gga tgt gaa gag cga aat gcg cag ggc atg tgt tcg ttg gca			1248
Leu Ala Gly Cys Glu Glu Arg Asn Ala Gln Gly Met Cys Ser Leu Ala			
405	410	415	
ggt ttt acg caa atc gtg aat gaa gca cgc ata ccg gcg tgc agt ttg			1296
Gly Phe Thr Gln Ile Val Asn Glu Ala Arg Ile Pro Ala Cys Ser Leu			
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aga tct cat cac cat cac cat cac taa			1323
Arg Ser His His His His His His			
435	440		

&lt;210&gt; 2

&lt;211&gt; 440

&lt;212&gt; PRT

&lt;213&gt; Escherichia coli

&lt;400&gt; 2

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Pro	Gln	Ser	Ala	Phe	Ala	Gln	Ser	Glu	Pro	Glu	Leu	Lys	Leu	Glu	Ser	20	25	30	
Val	Val	Ile	Val	Ser	Arg	His	Gly	Val	Arg	Ala	Pro	Thr	Lys	Ala	Thr	35	40	45	
Gln	Leu	Met	Gln	Asp	Val	Thr	Pro	Asp	Ala	Trp	Pro	Thr	Trp	Pro	Val	50	55	60	
Lys	Leu	Gly	Trp	Leu	Thr	Pro	Arg	Gly	Gly	Glu	Leu	Ile	Ala	Tyr	Leu	65	70	75	80
Gly	His	Tyr	Gln	Arg	Gln	Arg	Leu	Val	Ala	Asp	Gly	Leu	Leu	Ala	Lys	85	90	95	
Lys	Gly	Cys	Pro	Gln	Ser	Gly	Gln	Val	Ala	Ile	Ile	Ala	Asp	Val	Asp	100	105	110	
Glu	Arg	Thr	Arg	Lys	Thr	Gly	Glu	Ala	Phe	Ala	Ala	Gly	Leu	Ala	Pro	115	120	125	
Asp	Cys	Ala	Ile	Thr	Val	His	Thr	Gln	Ala	Asp	Thr	Ser	Ser	Pro	Asp	130	135	140	
Pro	Leu	Phe	Asn	Pro	Leu	Lys	Thr	Gly	Val	Cys	Gln	Leu	Asp	Asn	Ala	145	150	155	160
Asn	Val	Thr	Asp	Ala	Ile	Leu	Ser	Arg	Ala	Gly	Gly	Ser	Ile	Ala	Asp	165	170	175	
Phe	Thr	Gly	His	Arg	Gln	Thr	Ala	Phe	Arg	Glu	Leu	Glu	Arg	Val	Leu	180	185	190	
Asn	Phe	Pro	Gln	Ser	Asn	Leu	Cys	Leu	Lys	Arg	Glu	Lys	Gln	Asp	Glu	195	200	205	
Ser	Cys	Ser	Leu	Thr	Gln	Ala	Leu	Pro	Ser	Glu	Leu	Lys	Val	Ser	Ala	210	215	220	
Asp	Asn	Val	Ser	Leu	Thr	Gly	Ala	Val	Ser	Leu	Ala	Ser	Met	Leu	Thr	225	230	235	240
Glu	Ile	Phe	Leu	Leu	Gln	Gln	Ala	Gln	Gly	Met	Pro	Glu	Pro	Gly	Trp	245	250	255	
Gly	Arg	Ile	Thr	Asp	Ser	His	Gln	Trp	Asn	Thr	Leu	Leu	Ser	Leu	His	260	265	270	
Asn	Ala	Gln	Phe	Tyr	Leu	Leu	Gln	Arg	Thr	Pro	Glu	Val	Ala	Arg	Ser	275	280	285	
Arg	Ala	Thr	Pro	Leu	Leu	Asp	Leu	Ile	Met	Ala	Ala	Leu	Thr	Pro	His	290	295	300	
Pro	Pro	Gln	Lys	Gln	Ala	Tyr	Gly	Val	Thr	Leu	Pro	Thr	Ser	Val	Leu	305	310	315	320
Phe	Ile	Ala	Gly	His	Asp	Thr	Asn	Leu	Ala	Asn	Leu	Gly	Gly	Ala	Leu	325	330	335	
Glu	Leu	Asn	Trp	Thr	Leu	Pro	Gly	Gln	Pro	Asp	Asn	Thr	Pro	Pro	Gly	340	345	350	
Gly	Glu	Leu	Val	Phe	Glu	Arg	Trp	Arg	Arg	Leu	Ser	Asp	Asn	Ser	Gln	355	360	365	
Trp	Ile	Gln	Val	Ser	Leu	Val	Phe	Gln	Thr	Leu	Gln	Gln	Met	Arg	Asp	370	375	380	
Lys	Thr	Pro	Leu	Ser	Leu	Asn	Thr	Pro	Pro	Gly	Glu	Val	Lys	Leu	Thr	385	390	395	400
Leu	Ala	Gly	Cys	Glu	Glu	Arg	Asn	Ala	Gln	Gly	Met	Cys	Ser	Leu	Ala	405	410	415	
Gly	Phe	Thr	Gln	Ile	Val	Asn	Glu	Ala	Arg	Ile	Pro	Ala	Cys	Ser	Leu	420	425	430	
Arg	Ser	His	His	His	His	His	His									435	440		

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49

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 <213> Artificial Sequence

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<400> 4  
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33

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 <222> 403  
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 tgatgcggcg cattagcatc gcatcaggca atcaataatg tcagatatga aaagcggaaa 180  
 catatcgatg aaagcgatct taatcccatt tttatctctt ctgattccgt taaccccgca 240  
 atctgcattc gctcagagtg agccggagct gaagctggaa agtgtggtga ttgtcagtcg 300  
 tcatggtgtg cgtgctccaa ccaaggccac gcaactgatg caggatgtca cccagacgc 360  
 atggccaacc tggccggtaa aactgggtga gctgacaccg cgnggtggtg agctaategc 420  
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 ctgcccgcag tctggtcagg tcgcgattat tgctgatgtc gacgagcgta cccgtaaaac 540  
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 agatacgtcc agtcccgatc cgttatttaa tcctctaaaa actggcggtt gccaaactgga 660  
 taacgcgaac gtgactgacg cgatcctcag cagggcagga gggtaattg ctgactttac 720  
 cgggcatcgg caaacggcgt ttcgcgaact ggaacgggtg cttaattttc cgcaatcaaa 780  
 cttgtgcctt aaacgtgaga aacaggacga aagctgttca ttaacgcagg cattaccatc 840  
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 gctgacggag atattttctc tgcaacaagc acagggaatg ccggagccgg ggtggggaag 960  
 gatcaccgat tcacaccagt ggaacacctt gctaagtttg cataacgcgc aattttattt 1020  
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cctcttttggc	ggtgttcaag	ccaaaacgcg	caaccagcag	gctggtgccca	acagaacgcc	1800
ccacgaccgc	ggcatcactc	accgccagca	tcggcggcgt	atcgacaatc	accagatcgt	1860
aatggtcggt	cgcccatccc	agtaattgac	gcacccgatac	g		1901

&lt;210&gt; 6

&lt;211&gt; 1901

&lt;212&gt; DNA

&lt;213&gt; Escherichia coli

&lt;220&gt;

&lt;221&gt; misc\_feature

&lt;222&gt; 403

&lt;223&gt; n = A,T,C or G

&lt;400&gt; 6

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catatcgatg	aaagcgatct	taatcccatt	tttatctctt	ctgattccgt	taaccccgca	240
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gctgacggag	atattttctc	tgcaacaagc	acagggaaatg	ccggagccgg	ggtggggaag	960
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ccacgaccgc	ggcatcactc	accgccagca	tcggcggcgt	atcgacaatc	accagatcgt	1860
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&lt;210&gt; 7

&lt;211&gt; 1901

&lt;212&gt; DNA

&lt;213&gt; Escherichia coli

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 <222> (188)...(1483)

<221> misc\_feature  
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tgatgcggcg cattagcatc gcatcaggca atcaataatg tcagatatga aaagcggaaa	180
catatcg atg aaa gcg atc tta atc cca ttt tta tct ctt ctg att ccg	229
Met Lys Ala Ile Leu Ile Pro Phe Leu Ser Leu Leu Ile Pro	
1 5 10	
tta acc ccg caa tct gca ttc gct cag agt gag ccg gag ctg aag ctg	277
Leu Thr Pro Gln Ser Ala Phe Ala Gln Ser Glu Pro Glu Leu Lys Leu	
15 20 25 30	
gaa agt gtg gtg att gtc agt cgt cat ggt gtg cgt gct cca acc aag	325
Glu Ser Val Val Ile Val Ser Arg His Gly Val Arg Ala Pro Thr Lys	
35 40 45	
gcc acg caa ctg atg cag gat gtc acc cca gac gca tgg cca acc tgg	373
Ala Thr Gln Leu Met Gln Asp Val Thr Pro Asp Ala Trp Pro Thr Trp	
50 55 60	
ccg gta aaa ctg ggt tgg ctg aca ccg cgn ggt ggt gag cta atc gcc	421
Pro Val Lys Leu Gly Trp Leu Thr Pro Arg Gly Gly Glu Leu Ile Ala	
65 70 75	
tat ctc gga cat tac caa cgc cag cgt ctg gta gcc gac gga ttg ctg	469
Tyr Leu Gly His Tyr Gln Arg Gln Arg Leu Val Ala Asp Gly Leu Leu	
80 85 90	
gcg aaa aag ggc tgc ccg cag tct ggt cag gtc gcg att att gct gat	517
Ala Lys Lys Gly Cys Pro Gln Ser Gly Gln Val Ala Ile Ile Ala Asp	
95 100 105 110	
gtc gac gag cgt acc cgt aaa aca ggc gaa gcc ttc gcc gcc ggg ctg	565
Val Asp Glu Arg Thr Arg Lys Thr Gly Glu Ala Phe Ala Ala Gly Leu	
115 120 125	
gca cct gac tgt gca ata acc gta cat acc cag gca gat acg tcc agt	613
Ala Pro Asp Cys Ala Ile Thr Val His Thr Gln Ala Asp Thr Ser Ser	
130 135 140	
ccc gat ccg tta ttt aat cct cta aaa act ggc gtt tgc caa ctg gat	661
Pro Asp Pro Leu Phe Asn Pro Leu Lys Thr Gly Val Cys Gln Leu Asp	
145 150 155	
aac gcg aac gtg act gac gcg atc ctc agc agg gca gga ggg tca att	709
Asn Ala Asn Val Thr Asp Ala Ile Leu Ser Arg Ala Gly Gly Ser Ile	
160 165 170	

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gac gaa agc tgt tca tta acg cag gca tta cca tcg gaa ctc aag gtg Asp Glu Ser Cys Ser Leu Thr Gln Ala Leu Pro Ser Glu Leu Lys Val 210 215 220	853
agc gcc gac aat gtc tca tta acc ggt gcg gta agc ctc gca tca atg Ser Ala Asp Asn Val Ser Leu Thr Gly Ala Val Ser Leu Ala Ser Met 225 230 235	901
ctg acg gag ata ttt ctc ctg caa caa gca cag gga atg ccg gag ccg Leu Thr Glu Ile Phe Leu Leu Gln Gln Ala Gln Gly Met Pro Glu Pro 240 245 250	949
ggg tgg gga agg atc acc gat tca cac cag tgg aac acc ttg cta agt Gly Trp Gly Arg Ile Thr Asp Ser His Gln Trp Asn Thr Leu Leu Ser 255 260 265 270	997
ttg cat aac gcg caa ttt tat ttg cta caa cgc acg cca gag gtt gcc Leu His Asn Ala Gln Phe Tyr Leu Leu Gln Arg Thr Pro Glu Val Ala 275 280 285	1045
cgc agc cgc gcc acc ccg tta tta gat ttg atc aag aca gcg ttg acg Arg Ser Arg Ala Thr Pro Leu Leu Asp Leu Ile Lys Thr Ala Leu Thr 290 295 300	1093
ccc cat cca ccg caa aaa cag gcg tat ggt gtg aca tta ccc act tca Pro His Pro Pro Gln Lys Gln Ala Tyr Gly Val Thr Leu Pro Thr Ser 305 310 315	1141
gtg ctg ttt atc gcc gga cac gat act aat ctg gca aat ctc ggc ggc Val Leu Phe Ile Ala Gly His Asp Thr Asn Leu Ala Asn Leu Gly Gly 320 325 330	1189
gca ctg gag ctc aac tgg acg ctt ccc ggt cag ccg gat aac acg ccg Ala Leu Glu Leu Asn Trp Thr Leu Pro Gly Gln Pro Asp Asn Thr Pro 335 340 345 350	1237
cca ggt ggt gaa ctg gtg ttt gaa cgc tgg cgt cgg cta agc gat aac Pro Gly Gly Glu Leu Val Phe Glu Arg Trp Arg Arg Leu Ser Asp Asn 355 360 365	1285
agc cag tgg att cag gtt tcg ctg gtc ttc cag act tta cag cag atg Ser Gln Trp Ile Gln Val Ser Leu Val Phe Gln Thr Leu Gln Gln Met 370 375 380	1333
cgt gat aaa acg ccg ctg tca tta aat acg ccg ccc gga gag gtg aaa Arg Asp Lys Thr Pro Leu Ser Leu Asn Thr Pro Pro Gly Glu Val Lys 385 390 395	1381
ctg acc ctg gca gga tgt gaa gag cga aat gcg cag ggc atg tgt tcg	1429



Leu Thr Leu Ala Gly Cys Glu Glu Arg Asn Ala Gln Gly Met Cys Ser  
 400 405 410

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 Leu Ala Gly Phe Thr Gln Ile Val Asn Glu Ala Arg Ile Pro Ala Cys  
 415 420 425 430

agt ttg taatgcataa aaaagagcat tcagttacct gaatgctctg aggctgatga 1533  
 Ser Leu

caaacgaaga actgtctaat gcgtagaccg gaaaaggcgt tcacgccgca tccggccact 1593  
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 Gln Leu Met Gln Asp Val Thr Pro Asp Ala Trp Pro Thr Trp Pro Val  
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 Lys Leu Gly Trp Leu Thr Pro Arg Gly Gly Glu Leu Ile Ala Tyr Leu  
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 Gly His Tyr Gln Arg Gln Arg Leu Val Ala Asp Gly Leu Leu Ala Lys  
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 Lys Gly Cys Pro Gln Ser Gly Gln Val Ala Ile Ile Ala Asp Val Asp  
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 Glu Arg Thr Arg Lys Thr Gly Glu Ala Phe Ala Ala Gly Leu Ala Pro  
 115 120 125  
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 Phe Thr Gly His Arg Gln Thr Ala Phe Arg Glu Leu Glu Arg Val Leu  
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 Ser Cys Ser Leu Thr Gln Ala Leu Pro Ser Glu Leu Lys Val Ser Ala  
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 Asp Asn Val Ser Leu Thr Gly Ala Val Ser Leu Ala Ser Met Leu Thr  
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 Glu Ile Phe Leu Leu Gln Gln Ala Gln Gly Met Pro Glu Pro Gly Trp  
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Gly	Arg	Ile	Thr	Asp	Ser	His	Gln	Trp	Asn	Thr	Leu	Leu	Ser	Leu	His
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		275					280					285			
Arg	Ala	Thr	Pro	Leu	Leu	Asp	Leu	Ile	Lys	Thr	Ala	Leu	Thr	Pro	His
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Pro	Pro	Gln	Lys	Gln	Ala	Tyr	Gly	Val	Thr	Leu	Pro	Thr	Ser	Val	Leu
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Phe	Ile	Ala	Gly	His	Asp	Thr	Asn	Leu	Ala	Asn	Leu	Gly	Gly	Ala	Leu
			325						330					335	
Glu	Leu	Asn	Trp	Thr	Leu	Pro	Gly	Gln	Pro	Asp	Asn	Thr	Pro	Pro	Gly
			340					345					350		
Gly	Glu	Leu	Val	Phe	Glu	Arg	Trp	Arg	Arg	Leu	Ser	Asp	Asn	Ser	Gln
		355					360					365			
Trp	Ile	Gln	Val	Ser	Leu	Val	Phe	Gln	Thr	Leu	Gln	Gln	Met	Arg	Asp
	370					375					380				
Lys	Thr	Pro	Leu	Ser	Leu	Asn	Thr	Pro	Pro	Gly	Glu	Val	Lys	Leu	Thr
385					390					395					400
Leu	Ala	Gly	Cys	Glu	Glu	Arg	Asn	Ala	Gln	Gly	Met	Cys	Ser	Leu	Ala
			405						410					415	
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Pro	Gln	Ser	Ala	Phe	Ala	Gln	Ser	Glu	Pro	Glu	Leu	Lys	Leu	Glu	Ser	
			20					25					30			
gtg	gtg	att	gtc	agt	cgt	cat	ggc	gtg	cgt	gct	cca	acc	aag	gcc	acg	144
Val	Val	Ile	Val	Ser	Arg	His	Gly	Val	Arg	Ala	Pro	Thr	Lys	Ala	Thr	
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Gln	Leu	Met	Gln	Asp	Val	Thr	Pro	Asp	Ala	Trp	Pro	Thr	Trp	Pro	Val	
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Lys	Leu	Gly	Glu	Leu	Thr	Pro	Arg	Gly	Gly	Glu	Leu	Ile	Ala	Tyr	Leu	
65					70					75					80	
gga	cat	tac	tgg	cgt	cag	cgt	ctg	gta	gcc	gac	gga	tgg	ctg	cct	aaa	288
Gly	His	Tyr	Trp	Arg	Gln	Arg	Leu	Val	Ala	Asp	Gly	Leu	Leu	Pro	Lys	

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gac tgt gca ata acc gta cat acc cag gca gat acg tcc agt ccc gat Asp Cys Ala Ile Thr Val His Thr Gln Ala Asp Thr Ser Ser Pro Asp 130 135 140			432
ccg tta ttt aat cct cta aaa act ggc gtt tgc caa ctg gat aac gcg Pro Leu Phe Asn Pro Leu Lys Thr Gly Val Cys Gln Leu Asp Asn Ala 145 150 155 160			480
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aat ttt ccg caa tca aac ttg tgc ctt aaa cgt gag aaa cag gac gaa Asn Phe Pro Gln Ser Asn Leu Cys Leu Lys Arg Glu Lys Gln Asp Glu 195 200 205			624
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gag ata ttt ctc ctg caa caa gca cag gga atg ccg gag ccg ggg tgg Glu Ile Phe Leu Leu Gln Gln Ala Gln Gly Met Pro Glu Pro Gly Trp 245 250 255			768
gga agg atc acc gat tca cac cag tgg aac acc ttg cta agt ttg cat Gly Arg Ile Thr Asp Ser His Gln Trp Asn Thr Leu Leu Ser Leu His 260 265 270			816
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cgc gcc acc ccg tta tta gat ttg atc aag aca gcg ttg acg ccc cat Arg Ala Thr Pro Leu Leu Asp Leu Ile Lys Thr Ala Leu Thr Pro His 290 295 300			912
cca ccg caa aaa cag gcg tat ggt gtg aca tta ccc act tca gtg ctg Pro Pro Gln Lys Gln Ala Tyr Gly Val Thr Leu Pro Thr Ser Val Leu 305 310 315 320			960

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 325 330 335

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tgg att cag gtt tcg ctg gtc ttc cag act tta cag cag atg cgt gat 1152  
 Trp Ile Gln Val Ser Leu Val Phe Gln Thr Leu Gln Gln Met Arg Asp  
 370 375 380

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 385 390 395 400

ctg gca gga tgt gaa gag cga aat gcg cag ggc atg tgt tcg ttg gca 1248  
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 Val Val Ile Val Ser Arg His Gly Val Arg Ala Pro Thr Lys Ala Thr  
 35 40 45  
 Gln Leu Met Gln Asp Val Thr Pro Asp Ala Trp Pro Thr Trp Pro Val  
 50 55 60  
 Lys Leu Gly Glu Leu Thr Pro Arg Gly Gly Glu Leu Ile Ala Tyr Leu  
 65 70 75 80  
 Gly His Tyr Trp Arg Gln Arg Leu Val Ala Asp Gly Leu Leu Pro Lys  
 85 90 95  
 Cys Gly Cys Pro Gln Ser Gly Gln Val Ala Ile Ile Ala Asp Val Asp  
 100 105 110  
 Glu Arg Thr Arg Lys Thr Gly Glu Ala Phe Ala Ala Gly Leu Ala Pro  
 115 120 125  
 Asp Cys Ala Ile Thr Val His Thr Gln Ala Asp Thr Ser Ser Pro Asp

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Pro Leu Phe Asn Pro Leu Lys Thr Gly Val Cys Gln Leu Asp Asn Ala				
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Asn Val Thr Asp Ala Ile Leu Glu Arg Ala Gly Gly Ser Ile Ala Asp				160
	165		170	
Phe Thr Gly His Tyr Gln Thr Ala Phe Arg Glu Leu Glu Arg Val Leu				175
	180		185	
Asn Phe Pro Gln Ser Asn Leu Cys Leu Lys Arg Glu Lys Gln Asp Glu				190
	195		200	
Ser Cys Ser Leu Thr Gln Ala Leu Pro Ser Glu Leu Lys Val Ser Ala				205
	210		215	
Asp Cys Val Ser Leu Thr Gly Ala Val Ser Leu Ala Ser Met Leu Thr				220
225		230		235
Glu Ile Phe Leu Leu Gln Gln Ala Gln Gly Met Pro Glu Pro Gly Trp				240
	245		250	
Gly Arg Ile Thr Asp Ser His Gln Trp Asn Thr Leu Leu Ser Leu His				255
	260		265	
Asn Ala Gln Phe Asp Leu Leu Gln Arg Thr Pro Glu Val Ala Arg Ser				270
	275		280	
Arg Ala Thr Pro Leu Leu Asp Leu Ile Lys Thr Ala Leu Thr Pro His				285
	290		295	
Pro Pro Gln Lys Gln Ala Tyr Gly Val Thr Leu Pro Thr Ser Val Leu				300
305		310		315
Phe Ile Ala Gly His Asp Thr Asn Leu Ala Asn Leu Gly Gly Ala Leu				320
	325		330	
Glu Leu Asn Trp Thr Leu Pro Gly Gln Pro Asp Asn Thr Pro Pro Gly				335
	340		345	
Gly Glu Leu Val Phe Glu Arg Trp Arg Arg Leu Ser Asp Asn Ser Gln				350
	355		360	
Trp Ile Gln Val Ser Leu Val Phe Gln Thr Leu Gln Gln Met Arg Asp				365
	370		375	
Lys Thr Pro Leu Ser Leu Asn Thr Pro Pro Gly Glu Val Lys Leu Thr				380
385		390		395
Leu Ala Gly Cys Glu Glu Arg Asn Ala Gln Gly Met Cys Ser Leu Ala				400
	405		410	
Gly Phe Thr Gln Ile Val Asn Glu Ala Arg Ile Pro Ala Cys Ser Leu				415
	420		425	
				430



FIGURE 1a

(SEQ ID NO:1-nucleotide sequence and SEQ ID NO:2-amino acid sequence)

*Escherichia coli* B Phytase Sequence

1  
ATG AAA GCG ATC TTA ATC CCA TTT TTA TCT CTT CTG ATT CCG TTA ACC CCG  
Met Lys Ala Ile Leu Ile Pro Phe Leu Ser Leu Leu Ile Pro Leu Thr Pro  
CAA TCT GCA TTC GCT CAG AGT GAG CCG GAG CTG AAG CTG GAA AGT GTG GTG  
Gln Ser Ala Phe Ala Gln Ser Glu Pro Glu Leu Lys Leu Glu Ser Val Val  
ATT GTC AGT CGT CAT GGT GTG CGT GCT CCA ACC AAG GCC ACG CAA CTG ATG  
Ile Val Ser Arg His Gly Val Arg Ala Pro Thr Lys Ala Thr Gln Leu Met  
CAG GAT GTC ACC CCA GAC GCA TGG CCA ACC TGG CCG GTA AAA CTG GGT TGG  
Gln Asp Val Thr Pro Asp Ala Trp Pro Thr Trp Pro Val Lys Leu Gly Trp  
CTG ACA CCG CGN GGT GGT GAG CTA ATC GCC TAT CTC GGA CAT TAC CAA CGC  
Leu Thr Pro Arg Gly Gly Glu Leu Ile Ala Tyr Leu Gly His Tyr Gln Arg  
CAG CGT CTG GTA GCC GAC GGA TTG CTG GCG AAA AAG GGC TGC CCG CAG TCT  
Gln Arg Leu Val Ala Asp Gly Leu Leu Ala Lys Lys Gly Cys Pro Gln Ser  
GGT CAG GTC GCG ATT ATT GCT GAT GTC GAC GAG CGT ACC CGT AAA ACA GGC  
Gly Gln Val Ala Ile Ile Ala Asp Val Asp Glu Arg Thr Arg Lys Thr Gly  
GAA GCC TTC GCC GCC GGG CTG GCA CCT GAC TGT GCA ATA ACC GTA CAT ACC  
Glu Ala Phe Ala Ala Gly Leu Ala Pro Asp Cys Ala Ile Thr Val His Thr  
CAG GCA GAT ACG TCC AGT CCC GAT CCG TTA TTT AAT CCT CTA AAA ACT GGC  
Gln Ala Asp Thr Ser Ser Pro Asp Pro Leu Phe Asn Pro Leu Lys Thr Gly  
GTT TGC CAA CTG GAT AAC GCG AAC GTG ACT GAC GCG ATC CTC AGC AGG GCA  
Val Cys Gln Leu Asp Asn Ala Asn Val Thr Asp Ala Ile Leu Ser Arg Ala  
GGA GGG TCA ATT GCT GAC TTT ACC GGG CAT CGG CAA ACG GCG TTT CGC GAA  
Gly Gly Ser Ile Ala Asp Phe Thr Gly His Arg Gln Thr Ala Phe Arg Glu  
CTG GAA CGG GTG CTT AAT TTT CCG CAA TCA AAC TTG TGC CTT AAA CGT GAG  
Leu Glu Arg Val Leu Asn Phe Pro Gln Ser Asn Leu Cys Leu Lys Arg Glu  
AAA CAG GAC GAA AGC TGT TCA TTA ACG CAG GCA TTA CCA TCG GAA CTC AAG  
Lys Gln Asp Glu Ser Cys Ser Leu Thr Gln Ala Leu Pro Ser Glu Leu Lys  
GTG AGC GCC GAC AAT GTC TCA TTA ACC GGT GCG GTA AGC CTC GCA TCA ATG  
Val Ser Ala Asp Asn Val Ser Leu Thr Gly Ala Val Ser Leu Ala Ser Met  
CTG ACG GAG ATA TTT CTC CTG CAA CAA GCA CAG GGA ATG CCG GAG CCG GGG  
Leu Thr Glu Ile Phe Leu Leu Gln Gln Ala Gln Gly Met Pro Glu Pro Gly  
TGG GGA AGG ATC ACC GAT TCA CAC CAG TGG AAC ACC TTG CTA AGT TTG CAT  
Trp Gly Arg Ile Thr Asp Ser His Gln Trp Asn Thr Leu Leu Ser Leu His  
AAC GCG CAA TTT TAT TTG CTA CAA CGC ACG CCA GAG GTT GCC CGC AGC CGC  
Asn Ala Gln Phe Tyr Leu Leu Gln Arg Thr Pro Glu Val Ala Arg Ser Arg  
GCC ACC CCG TTA TTG GAT TTG ATC ATG GCA GCG TTG ACG CCC CAT CCA CCG  
Ala Thr Pro Leu Leu Asp Leu Ile Met Ala Ala Leu Thr Pro His Pro Pro



FIGURE 1b

CAA AAA CAG GCG TAT GGT GTG ACA TTA CCC ACT TCA GTA CTG TTT ATT GCC  
Gln Lys Gln Ala Tyr Gly Val Thr Leu Pro Thr Ser Val Leu Phe Ile Ala

GGA CAC GAT ACT AAT CTG GCA AAT CTC GGC GGC GCA CTG GAG CTC AAC TGG  
Gly His Asp Thr Asn Leu Ala Asn Leu Gly Gly Ala Leu Glu Leu Asn Trp

ACG CTT CCC GGT CAG CCG GAT AAC ACG CCG CCA GGT GGT GAA CTG GTG TTT  
Thr Leu Pro Gly Gln Pro Asp Asn Thr Pro Pro Gly Gly Glu Leu Val Phe

GAA CGC TGG CGT CGG CTA AGC GAT AAC AGC CAG TGG ATT CAG GTT TCG CTG  
Glu Arg Trp Arg Arg Leu Ser Asp Asn Ser Gln Trp Ile Gln Val Ser Leu

GTC TTC CAG ACT TTA CAG CAG ATG CGT GAT AAA ACG CCG CTG TCA TTA AAT  
Val Phe Gln Thr Leu Gln Gln Met Arg Asp Lys Thr Pro Leu Ser Leu Asn

ACG CCG CCC GGA GAG GTG AAA CTG ACC CTG GCA GGA TGT GAA GAG CGA AAT  
Thr Pro Pro Gly Glu Val Lys Leu Thr Leu Ala Gly Cys Glu Glu Arg Asn

GCG CAG GGC ATG TGT TCG TTG GCA GGT TTT ACG CAA ATC GTG AAT GAA GCA  
Ala Gln Gly Met Cys Ser Leu Ala Gly Phe Thr Gln Ile Val Asn Glu Ala

CGC ATA CCG GCG TGC AGT TTG AGA TCT CAT CAC CAT CAC CAT CAC TAA 1323  
Arg Ile Pro Ala Cys Ser Leu Arg Ser His His His His His His End



Amino acid sequence for E. coli appA (wild type) (SEQ ID NO:8)

MKAILIPFLSLLIPLTPQSAFAQSEPELKLESVVIVSRHGVRAPTKATQLMQDVTP  
DAWPTWPVKLGWLTPRGGELIAYLGHYQRQRLVADGLLAKKGCPQSGQVAIIA  
DVDERTRKTEGAFAAGLAPDCAITVHTQADTSSPDPLFNPLKTGVCQLDNA  
NVTDAILSRAGGSIADFTGHRQTAFRELERVLNFPQSNLCLKREKQDESCSLTQA  
LPSELKVSADNVSLTGAVSLASMLTEIFLLQQAQGMPEPGWGGRITDSHQWNTLL  
SLHNAQFYLLQRTPEVARSRATPLLDLIKALTTPHPQKQAYGVTLPSTVLFIAHG  
DTNLANLGGALELNWTLPGQPDNTPPGGELVFERWRRLSDNSQWIVSLVFQTL  
QQMRDKTPLSLNTPPGEVKLTLAGCEERNAQGMCSLAGFTQIVNEARIPACSL

Bold-Underlined amino acid residues are shown below in the modified appA enzyme  
(SEQ ID NO:10)

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ADVDERTRKTGEAFAAGLAPDCAITVHTQADTSSPDPLFNPLKTGVCQLDNA  
NVTDAILEAGGSIADFTGHYQTAFRELERVLNFPQSNLCLKREKQDESCSLTQAL  
PSELKVSADCVSLTGAVSLASMLTEIFLLQQAQGMPEPGWGGRITDSHQWNTLLS  
LHNAQFDLLQRTPEVARSRATPLLDLIKALTTPHPQKQAYGVTLPSTVLFIAHG  
DTNLANLGGALELNWTLPGQPDNTPPGGELVFERWRRLSDNSQWIVSLVFQTL  
QQMRDKTPLSLNTPPGEVKLTLAGCEERNAQGMCSLAGFTQIVNEARIPACSLRS  
~~HL~~

Figure 8